Supporting Information

S-Nitroso-*N*-acetylpenicillamine (SNAP) impregnated silicone Foley catheters: a potential biomaterial/device to prevent catheter associated urinary tract infections

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Biofilm Growth Conditions and Plate Counting

***S. epidermidis* biofilm.** *S. epidermidis* ATCC 14990 was tested as a bacterial strain to develop 3, 7, and 14 day-old biofilms on the surfaces of both control and SNAP impregnated silicone Foley catheter segments. A CDC biofilm reactor (Biosurface Technologies, Bozeman, MT) was used for the biofilm growth, and the CDC biofilm reactor and its coupon holders were autoclaved before use. Both control and SNAP impregnated catheters segments were mounted on the coupon holders and the reactor was supplemented with 10% LB medium by a peristaltic pump with a continuous flow rate of 100 mL/h. Overnight cultures of *S. epidermidis* (grown under shaking conditions at 37 °C) were diluted by 1:100 and inoculated into the glass vessel of the CDC reactor aseptically. The liquid growth medium was circulated through the vessel and a magnetic stir bar rotated by a magnetic stir plate was used to generate a shear force. The CDC biofilm reactor was placed inside an incubator and biofilms were grown at 37 °C. After 3, 7 or 14 d of growth, the catheters were aseptically removed and each catheter was cut into two 1 cm pieces. One piece was utilized for fluorescent imaging of live bacteria on the surface, while the other 1 cm segment was used for plate count experiments. Each 1 cm segment to be used for plate counting was placed into a centrifuge tube, containing 2 mL of 10 mM sterilized PBS (pH 7.4). The catheter piece was then homogenized for 30 s in order to disintegrate the biofilm clumps and form a homogeneous single cell suspension. Finally, to assess cell viability, samples were serially diluted by 10-fold each time and plated onto LB agar plates. All experiments were conducted in triplicate.

***P. mirabilis* biofilm.** The bacterium *P. mirabilis* ATCC 29906 was also tested for the development of 3, 7, and 14 day-old biofilms on the surfaces of both control and SNAP impregnated silicone Foley catheter segments. Growth conditions were the same as those used for *S. epidermidis* biofilms. Procedures to assess cell viability were also performed exactly as described above for *S. epidermidis*. For the second studies with *P. mirabilis*, the SNAP catheter segments were presoaked for 24 h before placement in the bioreactor. All experiments were conducted in triplicate.

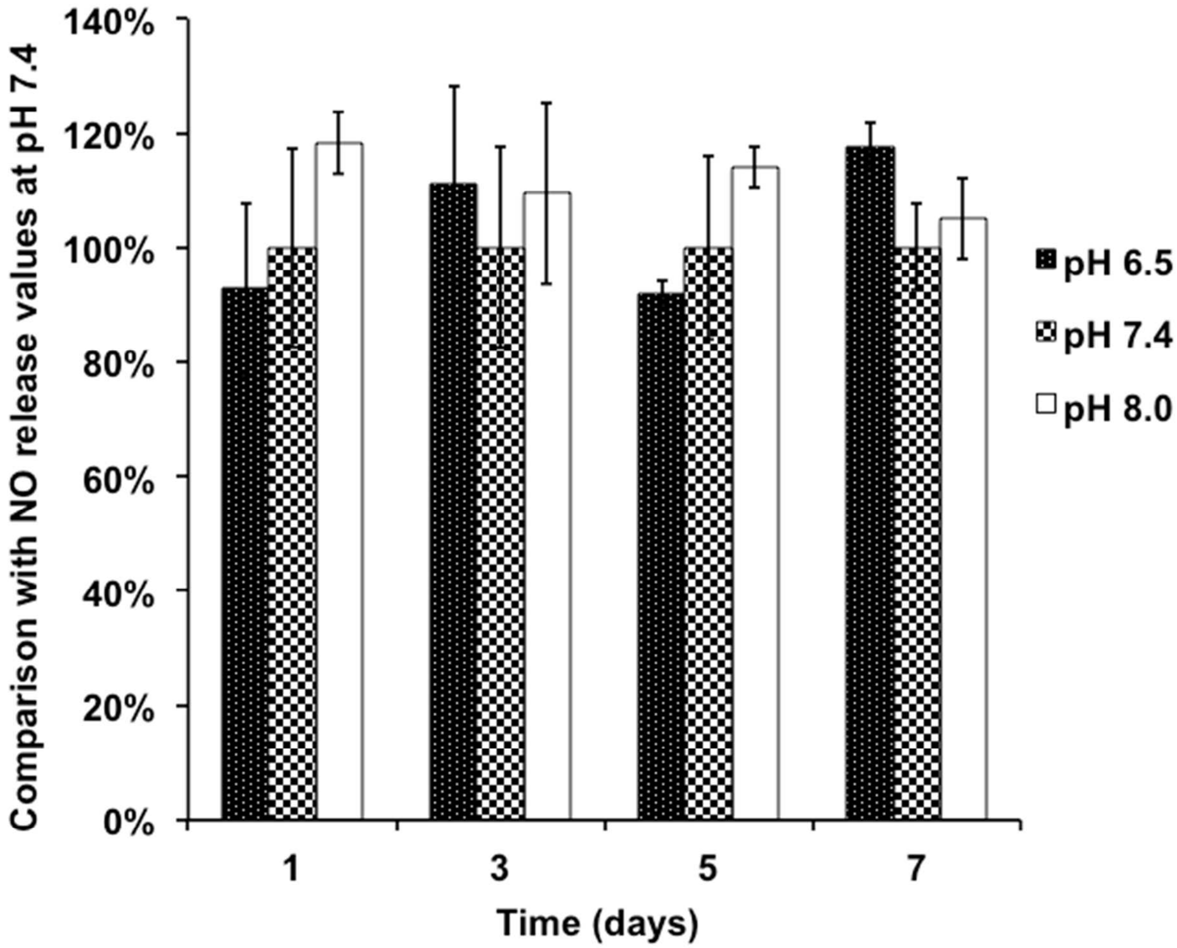

Figure S1. Comparison of measured NO release fluxes from SNAP impregnated Foley catheter tubing soaked for 7 d in different pH PBS solutions (pH 6.5, 7.4 and 8.0) at 37 ^{o}C and tested every other day for NO release rates under these same conditions with fresh buffers of the same pH values. Data (n = 3 pieces of tubing for each condition) normalized to NO fluxes observed on each test day measured at pH 7.4 (100%) ± SEM.

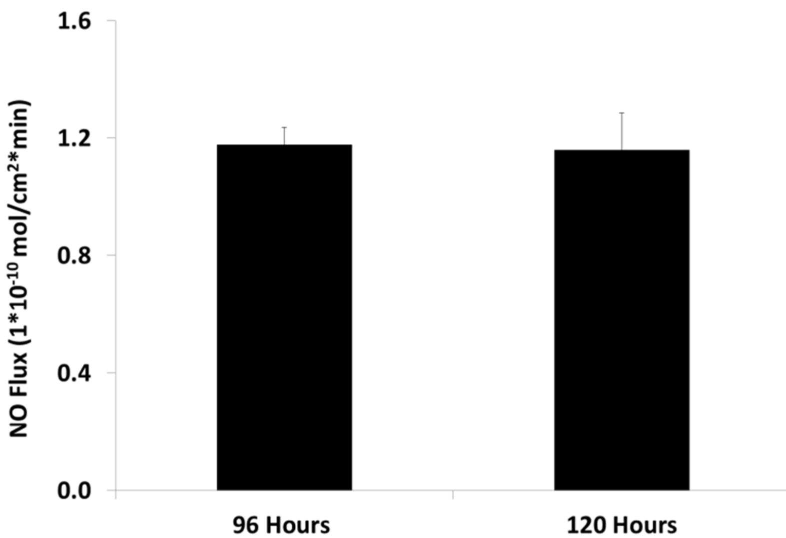

Figure S2. Nitric oxide flux of the SNAP impregnated catheter segments after 3 d *P. mirabilis* biofilm experiment in a CDC biofilm reactor (n=3). These catheter pieces had been pre-soaked in PBS buffer for 24 h prior to the start of the experiment. 96 h and 120 h time-points, respectively, correspond to the day of cell viability assessment and the following day.